



Redescription and Molecular Characterisation of the Fish Parasitic Isopod *Norileca indica* (Milne Edwards, 1840) (Crustacea: Isopoda: Cymothoidae) with a Key to the Genus

Authors: Wal, Serita van der, Smit, Nico J, and Hadfield, Kerry A

Source: African Zoology, 52(3) : 163-175

Published By: Zoological Society of Southern Africa

URL: <https://doi.org/10.1080/15627020.2017.1382389>

BioOne Complete (complete.BioOne.org) is a full-text database of 200 subscribed and open-access titles in the biological, ecological, and environmental sciences published by nonprofit societies, associations, museums, institutions, and presses.

Your use of this PDF, the BioOne Complete website, and all posted and associated content indicates your acceptance of BioOne's Terms of Use, available at www.bioone.org/terms-of-use.

Usage of BioOne Complete content is strictly limited to personal, educational, and non - commercial use. Commercial inquiries or rights and permissions requests should be directed to the individual publisher as copyright holder.

BioOne sees sustainable scholarly publishing as an inherently collaborative enterprise connecting authors, nonprofit publishers, academic institutions, research libraries, and research funders in the common goal of maximizing access to critical research.

Redescription and molecular characterisation of the fish parasitic isopod *Norileca indica* (Milne Edwards, 1840) (Crustacea: Isopoda: Cymothoidae) with a key to the genus

Serita van der Wal¹, Nico J Smit¹ and Kerry A Hadfield^{1,2*}

¹ Water Research Group, Unit for Environmental Sciences and Management, Potchefstroom Campus, North-West University, Potchefstroom, South Africa

² Department of Zoology and Entomology, University of the Free State, Bloemfontein, South Africa

* Corresponding author, email: kerryh26@yahoo.com

***Norileca indica* (Milne Edwards, 1840) is fully redescribed based on ovigerous females collected from Maputo Bay, Mozambique, from the branchial cavity of the fish host *Selar crumenophthalmus* Bloch, 1793. An identification key to the species of *Norileca* Bruce, 1990 is given. Furthermore, a fragment of the mitochondrial cytochrome oxidase I (COI) gene from *N. indica* was sequenced for the first time. This is the first molecular characterisation of a species of *Norileca*.**

Keywords: branchial cavity, cymothoid, genetic characterisation, *Norileca*, *Selar crumenophthalmus*, taxonomy

Introduction

Species of *Norileca* Bruce, 1990 inhabit the branchial cavity of fish hosts and are commonly recorded from pelagic fishes (Rameshkumar et al. 2015). There are three known species: *Norileca borealis* Javed & Yasmeen, 1999, *N. triangulata* Richardson, 1910 and *N. indica* (Milne Edwards, 1840). *Norileca borealis* was originally described from the northern Arabian sea (Javed and Yasmeen 1999), parasitising the Indian mackerel, *Rastrelliger kanagurta* Cuvier, 1817. No other recordings of this species have been reported since its original description. *Norileca triangulata* was first recorded from Tanimdao Island, the Philippines, without mention of a fish host (Richardson 1910). Specimens of *N. triangulata* have since been recorded from Australia (from Cape York, Great Barrier Reef and south-eastern Queensland), from the branchial cavity of the sailfin flyingfish, *Parexocoetus brachypterus* Richardson, 1846 and *Sardinella gibbosa* Bleeker, 1849 (Bruce 1990). More recent recordings are from the Parangipettai coastal waters on the south-east coast of India, from the goldstripe sardinella, *Sardinella gibbosa* Bleeker, 1849 (Rameshkumar and Ravichandran 2015). *Norileca indica* was originally described by Milne Edwards (1840) as *Livoneca indica*. It was later redescribed by Bruce (1990) and transferred to the genus *Norileca*.

As part of a larger project on the global diversity of cymothoids, *N. indica* specimens were collected from Maputo Bay, Mozambique, and provided the opportunity to complete a redescription of this species based on ovigerous females. In addition, this paper also presents a detailed redescription of the male, as well as the first molecular characterisation of this species and genus, using the mitochondrial cytochrome oxidase I (COI) gene.

Methods

Norileca indica specimens were collected during November 2013 from the bigeye scad, *Selar crumenophthalmus* Bloch 1793 by local subsistence fishermen in Maputo Bay, Mozambique. Isopods were analysed following the techniques of Hadfield et al. (2010, 2013). Species descriptions were made with the aid of the taxonomy software package DELTA (Descriptive Language for Taxonomy) (see Coleman et al. 2010), following a general Cymothoidae character data set originally developed by Hadfield et al. (2013) and recently updated (Hadfield et al. 2016). Ratios and measurements for the description were made using the maximum values at the middle of the specific measured article, and all proportional measurements were rounded to one decimal place. Isopod classification follows Brandt and Poore (2003) and host nomenclature follows that of FishBase (Froese and Pauly 2017) and *Catalog of Fishes* (Eschmeyer et al. 2017).

Genomic DNA was extracted from isopod pereopods and pleopods following the protocol for animal tissue extraction of the GeneJET™ Genomic Extraction Kit (Thermo Scientific, Waltham, MA, USA). A targeted part of the mitochondrial cytochrome oxidase I (COI) gene (approximately 680 bp) of these specimens was subjected to PCR amplification with the aid of a Bio-Rad C1000 Touch™ Thermal Cycler and universal invertebrate primers LCO1490 (5'-GGTCAACAATCATAAAGATATTGG-3') and HC02198 (5'-TAAACTTCAGGGTGACCAAAAAATCA-3') (Folmer et al. 1994). The PCR protocol followed that of Ketmaier et al. (2008). PCR products were sequenced in both directions by Inqaba Biotechnical Industries

(Pty) Ltd, Pretoria, South Africa. The bioinformatics software platform, Geneious R9.1 (Biomatters, Auckland, New Zealand; available at <http://www.geneious.com>), was used to trim and align consensus sequences. Sequences were deposited into the NCBI GenBank database (<http://www.ncbi.nlm.nih.gov/genbank/>) with the following accession numbers: MF628258, MF628259 and MF628260.

Abbreviations

MNHN = National Museum of Natural History, Paris, France; NWU = North-West University, Potchefstroom Campus; SAM = Iziko South African Museum, Cape Town; TL = total length; W = width.

Taxonomy

Suborder: Cymothoida Wägele, 1989

Superfamily: Cymothoidea Leach, 1814

Family: Cymothoidae Leach, 1814

Genus *Norileca* Bruce, 1990

Norileca Bruce, 1990: 289.—Bruce, Lew Ton & Poore 2002: 181.

Type species: *Livoneca indica* Milne Edwards, 1840, by original designation (Bruce 1990).

Diagnosis

Body weakly vaulted dorsally, twisted to one side. Cephalon posterior margin medially indented, weakly or not trilobed. Coxae narrow, shorter than, or as long as respective pereonites. Brood pouch with 4 pairs of alternately overlapping oostegites on coxae 2–5. Pleon not immersed in pereon; pleonite 1 widest; pleonites 1–5 becoming progressively narrower; pleonites 1 and 2 without ventro-lateral processes. Uropods not extending beyond posterior margin of pleotelson. Pleopods 1–4 with laminar rami; peduncle articles 2–5 lateral margin with laminar lobe; endopods 3–5 folded proximomedial lobe present; endopod 5 distal margin medially indented, including 2 folded lobes. Pereopods lacking expanded lobes; without expanded carina on bases. Antennula shorter than, or subequal to antenna; bases of antennula wide apart. Mandible palp article 2 flattened, prominently expanded. Maxilliped lacking oostegital lobes.

Remarks

Norileca shares several characters with *Livoneca* Leach, 1818. Both genera are similar with regard to their pereopod morphology, all of which are robust and lacking an expanded carina on the base of the pereopods; the cephalon posterior margin is trilobed; and the pleon is not immersed in the pereon with pleonites 1–5 becoming progressively narrower. *Norileca* can be distinguished from *Livoneca* in having a weakly trilobed cephalon (vs strongly trilobed in *Livoneca*) and pleonites 1–3 lateral margins which are not bilobed. *Norileca* also has an expanded mandible palp article 3, pleopods 3–4 without folds on endopods, as well as an absence of branched pleopod

peduncles (Bruce 1990). It can be distinguished from other cymothoid genera by pleonite 1 being the widest of the pleonites, as well as its weakly twisted body shape (Hadfield 2012).

Key to the species of *Norileca*

1. Pleonite 5 narrower than pleonite 1; uropods almost reaching posterior margin of pleotelson; pleotelson approximately 0.9 times as long as wide **2**
Pleonite 5 and pleonite 1 subequal; uropods two-thirds the length of pleotelson; pleotelson approximately 1.0–1.2 times as long as wide ***N. indica***
2. Body twisted to the side; maxilla medial lobe with 1 robust seta and lateral lobe with 4 robust setae
..... ***N. borealis***
Body nearly straight; maxilla medial lobe with 2 robust setae and lateral lobe with 2 robust setae
..... ***N. triangulata***

Norileca indica Milne Edwards, 1840

Livoneca indica Milne Edwards, 1840: 262.—Bleeker, 1857: 21, 28.—Gerstaecker, 1882: 261.—Schioedte & Meinert, 1884: 362–365, pl. 5, figs. 3–6; Richardson, 1910: 24.—Nierstrasz, 1915: 99–100.—Nierstrasz, 1931: 142–143, 145.—Borcea, 1933: 482.—Beumer, Ashburner, Burbury, Jette & Latham, 1982: 33.

Livoneca ornata Heller, 1868: 145–146, pl. 12, fig. 15.—Gerstaecker, 1882: 261.

Lironeca indica.—Trilles, 1976: 777–778, pl. 2, fig. 3.—Avdeev, 1978: 281–282.—Trilles, 1979: 266.—Rokicki, 1982: 205–208, figs. 1–2.—Trilles, 1994: 178–179.

Norileca indica.—Bruce, 1990: 291–293.—Bruce, Lew Ton & Poore, 2002: 181.—Ghani, 2003: 219.—Yu & Li, 2003: 235–237, fig. 10.—Yamauchi, Ohtsuka & Nagasawa, 2005: 25–27.—Nagasawa & Petchsupa, 2009: 131–133.—Rameshkumar, Ravichandran & Sivasubramanian, 2013a: 99–105.—Rameshkumar, Ravichandran, Sivasubramanian & Trilles, 2013b: 42–46.—Argente, Narido, Palla & Celedonio, 2014: 3–8.—Neeraja, Tripathi & Shameem, 2014: 49–56.—Rameshkumar & Ravichandran, 2015: 33–36.—Rameshkumar, Ramesh, Ravichandran, Trilles & Subbiah, 2015: 712–715.—Aneesh, Kappalli, Kottathil, Gopinathan & Trilles, 2015: 42.—Behera, Ghosh & Pattnaik, 2016: 856–862.—Jithin, Swapna, Kumar, Venu, Helna & Sudha, 2016: 47–53.—Cruz-Laciera & Nagasawa, 2017: 60–63.

Type material: Holotype held at the Museum Nationale d'histoire Naturelle, Paris (MNHN-IU-2007-4159).

Type locality: Sumatra Island, Indonesia (Milne Edwards 1840).

Type host: No type host recorded.

Material examined

Three ovigerous ♀ (33.0 mm TL, 16 mm W; 30.0 mm TL, 17 mm W; 26.0 mm TL, 13 mm W) and 1 ♂ (11.0 mm TL; 3.0 mm W), Maputo Bay, Mozambique, South Africa, November 2013, from bigeye scad *Selar crumenophthalmus* (Bloch, 1793), coll. Wynand Vlok (SAMC-A089028).

Two ovigerous ♀ (29.0–35.0 mm TL; 15.0–19.0 mm W) and 2 non-ovigerous ♀ (28.0–30.0 mm TL; 14.0–18.0 mm W), Maputo Bay, Mozambique, South Africa, November 2013, from bigeye scad *Selar crumenophthalmus* (Bloch, 1793), coll. Wynand Vlok (in the collection of the authors at NWU).

Ovigerous female
(Figures 1–4)

Length 33.0 mm, width 16.0 mm.

Body twisted to the right side, 2.2 times as long as greatest width, dorsal surfaces smooth and polished in appearance, widest at pereonite 4, most narrow at pereonite 1. Pereonite lateral margins posteriorly protruding. *Cephalon* 1.1 times longer than wide, visible from dorsal view, triangular. *Frontal margin* thickened and ventrally folded. *Eyes* oval with distinct margins, one eye 0.3 times the width of the cephalon, 0.3 times the length of the cephalon. *Pereonite 1* smooth, with anterior border indented and anterolateral angle weakly produced, extending to middle of cephalon. *Coxae* 2–3

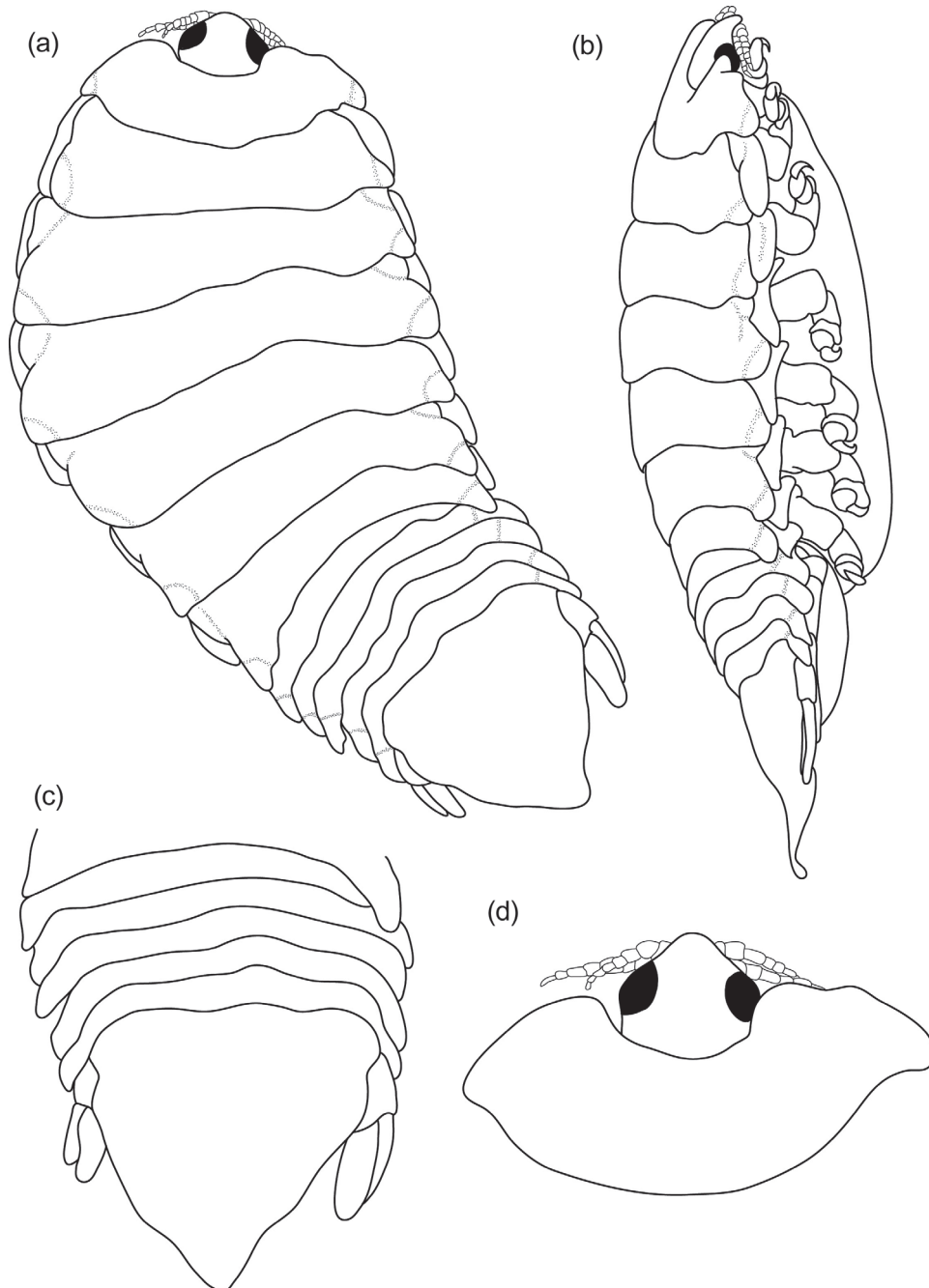


Figure 1: *Norileca indica* (Milne Edwards, 1840) ovigerous female (33.0 mm TL, 16.0 mm W) (SAMC-A089028). (a) Dorsal body, (b) lateral body, (c) dorsal view of pleotelson with uropods, (d) dorsal view of cephalon and pereonite 1

wide with posteroventral angles rounded; coxae 4–7 acute, posteriorly pointed, not extending past pereonite margin. Pereonites 6 and 7 narrower than pereonites 1–5. *Pleon* with pleonite 1 slightly wider than other pleonites, visible in dorsal view; pleonites posterior margin not smooth, medially produced. Pleonite 2 partially overlapped by pereonite 7 posterolateral margin; posterolateral angles of pleonite 2 rounded. Pleonites 3–5 similar in form to pleonite 2; pleonite 5 free, not overlapped by lateral margins of pleonite 4. *Pleotelson* as long as anterior width; dorsal surface smooth; lateral margins weakly convex, posteriorly narrow; posterior margin converging to caudomedial point.

Antennula consists of 8 articles; peduncle articles 1 and 2 distinct and articulated; article 2 1.1 times as long as article 1; article 3 1.6 times as long as wide, 0.5 times as long as combined lengths of articles 1 and 2; flagellum with 5 articles, extending to posterior margin of eye with tufts of simple setae on articles 3–6 and 8. *Antenna* consists of 9 articles; peduncle article 3 1.0 times as long as article 2; article 4 2.2 times as long as wide, 1.4 times as long as article 3; article 5 twice as long as wide, 0.7 times as long as article 4. Antenna flagellum with 6 articles, terminal article with 1–5 short simple setae, extending to anterior margin of pereonite 1. *Mandibular molar process* present, with no simple setae; mandible

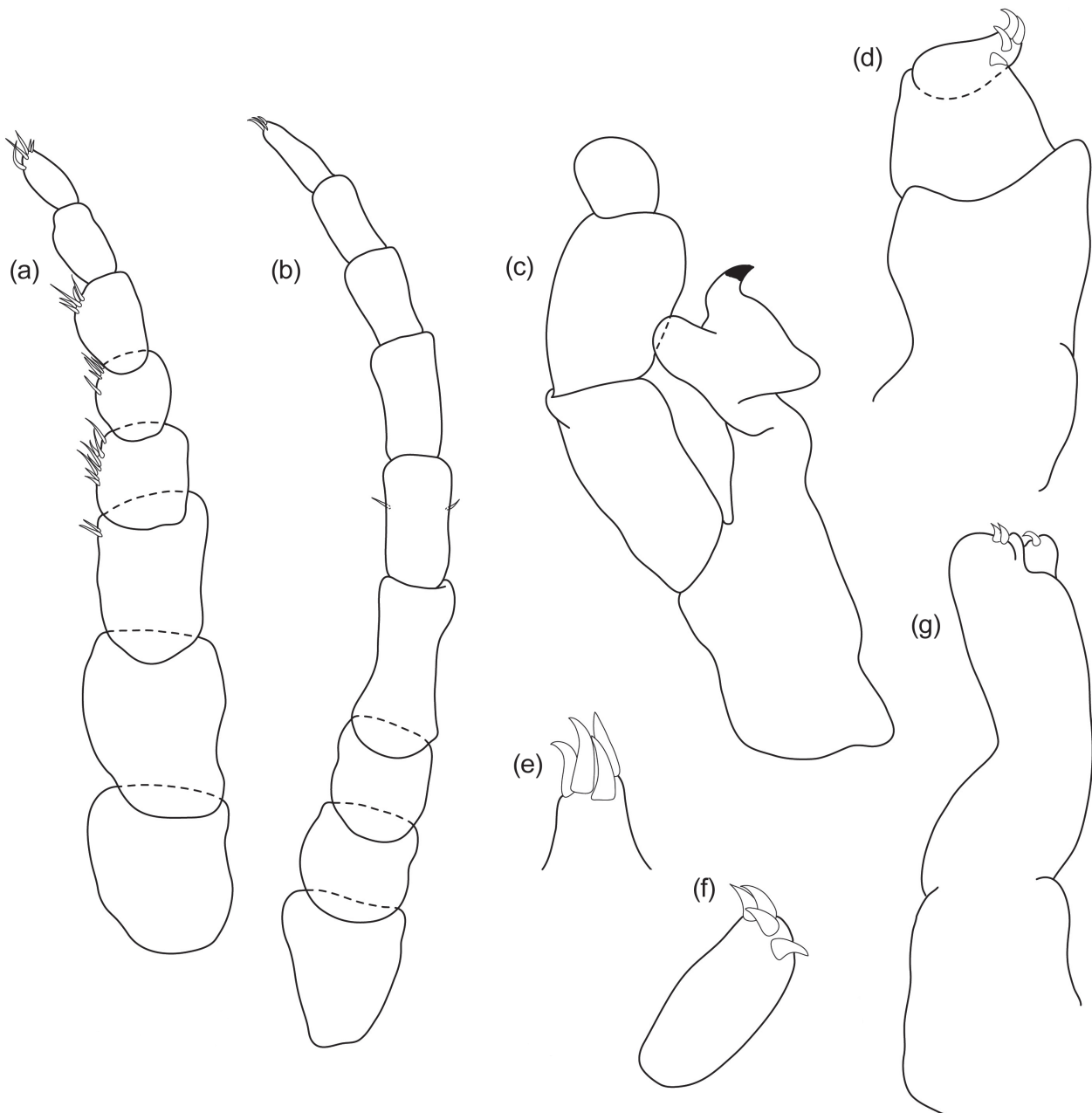


Figure 2: *Norileca indica* (Milne Edwards, 1840) ovigerous female (33.0 mm TL, 16.0 mm W) (SAMC-A089028). (a) Antennula, (b) antenna, (c) mandible, (d) maxilliped, (e) tip of maxillule, (f) tip of maxilliped article 3, (g) maxilla

palp article 2 and 3 without setae. *Maxillule* simple with 4 terminal robust setae. *Maxilla* medial lobe partly fused to lateral lobe; medial lobe with 2 recurved robust setae, lateral lobe with 1 large recurved robust setae. *Maxilliped* palp article 2 without setae; article 3 with 4 recurved robust setae.

Pereopod 1 basis 1.8 times as long as greatest width; ischium 0.7 times as long as basis; merus proximal margin with bulbous protrusion; carpus with straight proximal margin; propodus 1.1 times as long as wide; dactylus slender, 3.8 as long as propodus, 3.8 times as long as basal width. *Pereopods 3–6* similar to pereopod 2, gradually increasing in size towards posterior, all without setae. *Pereopod 7* basis 0.6 times as long as greatest width; ischium 0.8 as long as basis, without protrusions;

merus proximal margin with large bulbous protrusion; 0.4 times as long as wide, 0.3 as long as ischium; carpus 0.4 times as long as wide, 0.2 as long as ischium, with slight bulbous protrusion; propodus as long as wide, 0.4 as long as ischium; dactylus slender, 1.9 as long as propodus, 2.7 times as long as basal width.

Pleopods without setae, exopod larger than endopod. *Pleopod 1* exopod 1.1 times as long as wide, lateral margin weakly convex, distally broadly rounded, medial margin strongly convex; endopod 1.2 times as long as wide, lateral margin convex, distally broadly rounded, medial margin straight; peduncle 0.3 times as wide as long. *Pleopods 2–5* similar to pleopod 1. *Pleopods 3–5* with fleshy folds present, increasing in size from pleopod 3–5. Peduncle lobes present, increasing in size from pleopod 1–5.

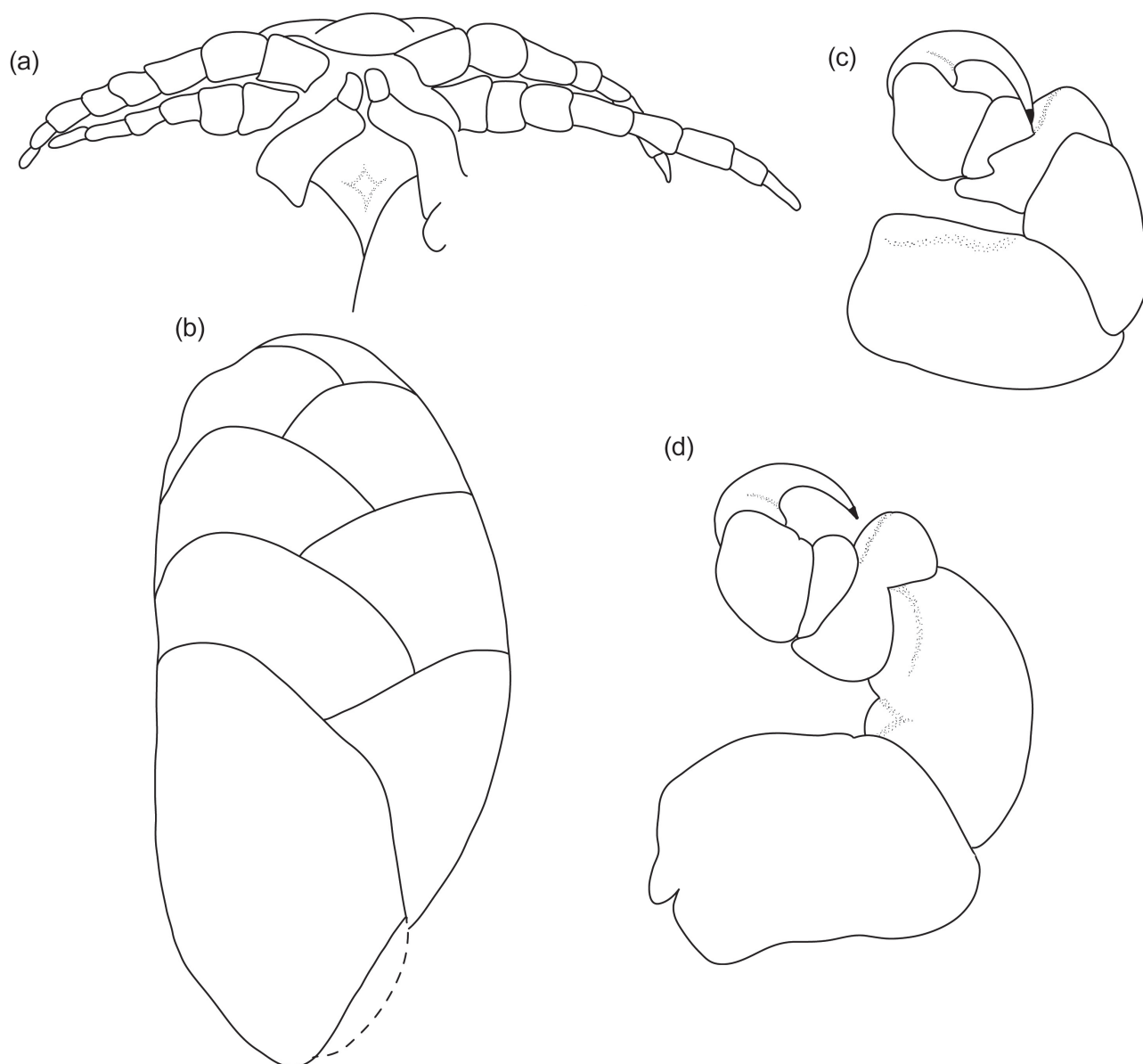


Figure 3: *Norileca indica* (Milne Edwards, 1840) ovigerous female (33.0 mm TL, 16.0 mm W) (SAMC-A089028). (a) Ventral cephalon, (b) oostegites, (c) pereopod 1, (d) pereopod 7

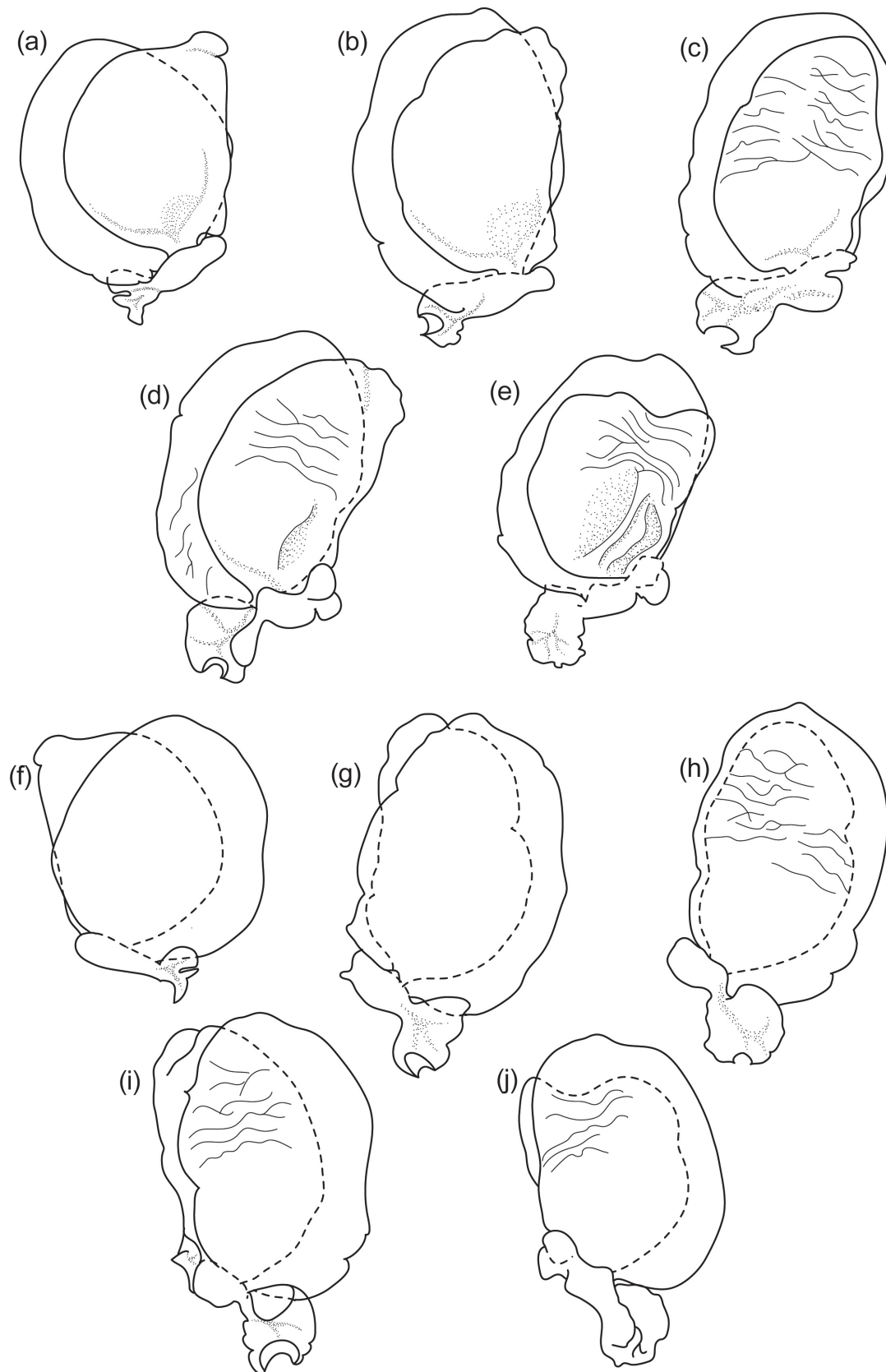


Figure 4: *Norileca indica* (Milne Edwards, 1840) ovigerous female (33.0 mm TL, 16.0 mm W) (SAMC-A089028). (a) Pleopod 1 dorsal view, (b) pleopod 2 dorsal view, (c) pleopod 3 dorsal view, (d) pleopod 4 dorsal view, (e) pleopod 5 dorsal view, (f) pleopod 1 ventral view, (g) pleopod 2 ventral view, (h) pleopod 3 ventral view, (i) pleopod 4 ventral view, (j) pleopod 5 ventral view

Uropod more than half the length of pleotelson; peduncle 0.7 times longer than rami, lateral margin without setae; rami not extending beyond pleotelson, marginal setae absent, apices narrowly rounded. *Endopod* 2.3 times as long as greatest width, without setae. *Exopod* not extending to end of endopod, 3 times as long as greatest width, without setae.

Male

(Figures 5–7)

Length 11.0 mm, width 3.0 mm.

Body straight, not twisted, 2.7 times as long as greatest width, widest at pereonite 5, most narrow at pereonite 1, pereonite lateral margins mostly posteriorly ovate. *Cephalon* 0.79 times longer than wide, visible from dorsal view, triangular, not immersed in pereonite 1. *Frontal margin* thickened, ventrally folded. *Eyes* oval with distinct margins; one eye 0.6 times length of cephalon. *Pereonite 1* smooth, anterior border indented; anterolateral angle weakly produced, extending past the posterior margin of

eyes. Posterior margins of pereonites smooth and straight. Coxae 2–3 wide; with posteroventral angles rounded; 4–7 acute, posteriorly pointed; not extending past pereonite margin. Pereonites 6 and 7 becoming more progressively rounded posteriorly. *Pleon* with pleonite 1 largely concealed by pereonite 7; pleonites posterior margin smooth, mostly concave. Pleonite 2 not overlapped by pereonite 7; postero-lateral angles of pleonite 2 rounded. Pleonites 3–5 similar in form to pleonite 2; pleonite 5 free, not overlapped by lateral margins of pleonite 4. *Pleotelson* as long as anterior width; dorsal surface smooth; lateral margins weakly convex, posterior margin converging to caudomedial point.

Antennula more stout than antenna, longer than antenna; consists of 8 articles; peduncle articles 1 and 2 distinct and articulated; article 2 as long as article 1; article 3 1.4 times as long as wide, 0.5 times as long as combined lengths of articles 1 and 2; flagellum with 5 articles, extending to anterior of pereonite 1, with tufts of setae on articles 3–8. *Antenna* consists of 9 articles; peduncle article 3 1.1 times as long as article 2; article 4 1.5 times as long as wide,

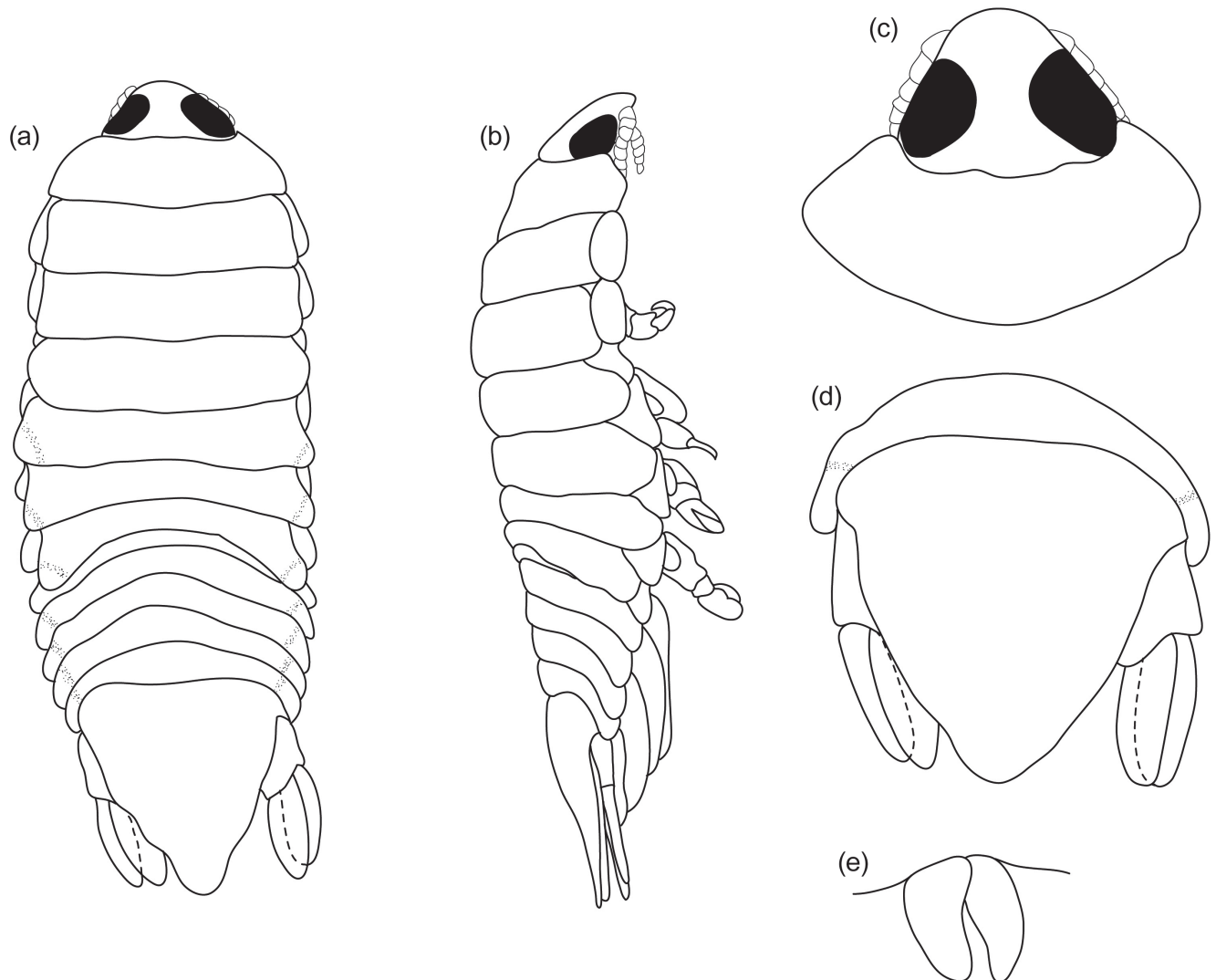


Figure 5: *Norileca indica* (Milne Edwards, 1840) male (11.0 mm TL, 3.0 mm W) (SAMC-A089028). (a) Dorsal body, (b) lateral body, (c) dorsal view of cephalon and pereonite 1, (d) dorsal view of pleotelson with uropods, (e) penes

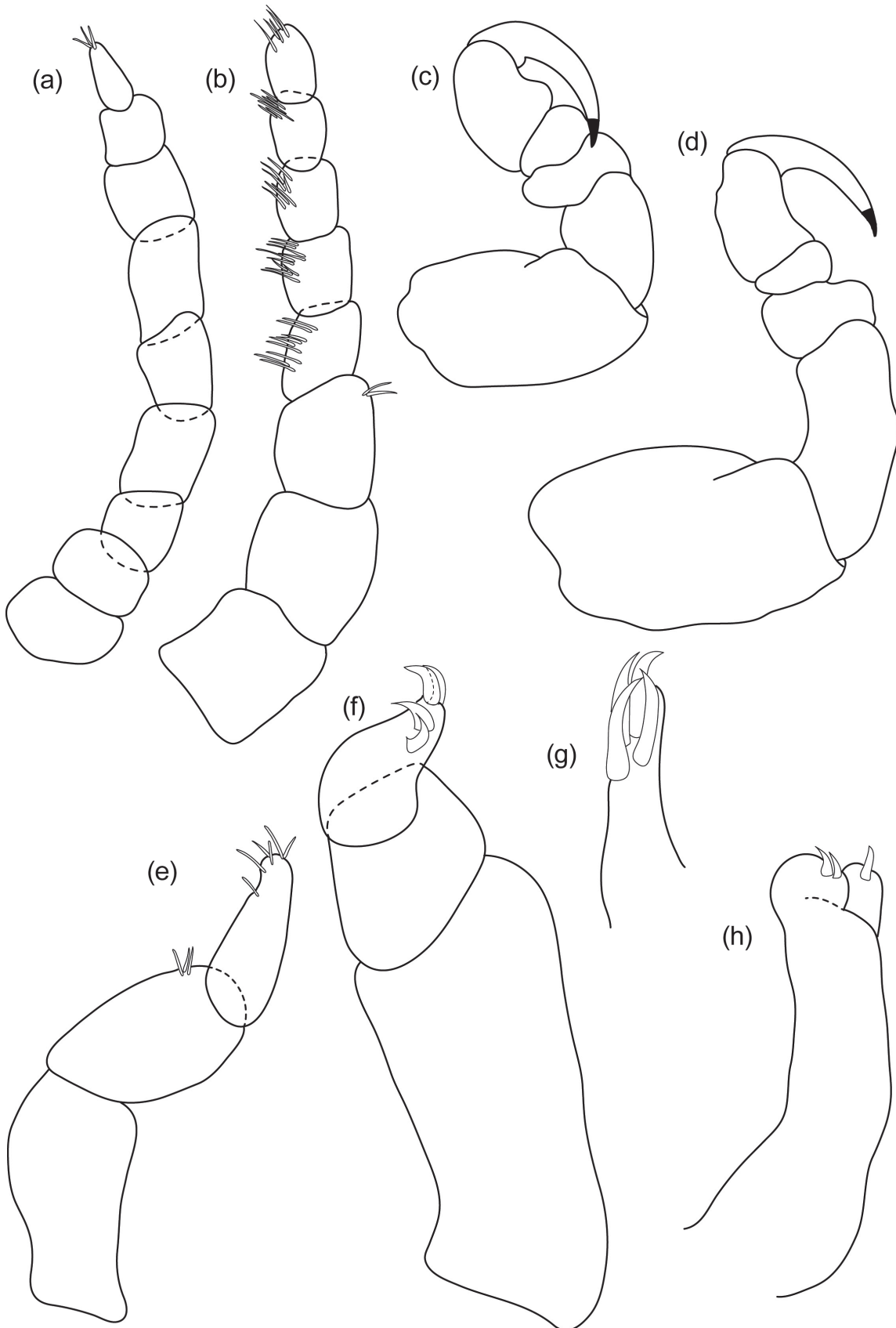


Figure 6: *Norileca indica* (Milne Edwards, 1840) male (11.0 mm TL, 3.0 mm W) (SAMC-A089028). (a) Antenna, (b) antennula, (c) pereopod 1, (d) pereopod 7, (e) mandibular palp, (f) maxilliped, (g) tip of maxillule, (h) maxilla

1.3 times as long as article 3; article 5 1.6 times as long as wide, as long as article 4. Antenna flagellum with 7 articles, terminal article terminating in 1–5 short simple setae, extending to anterior margin of pereonite 1. *Mandibular molar process* present; palp article 2 with 3 distolateral setae, article 3 with 6 simple setae. *Maxillule* simple with 4 terminal robust setae. *Maxilla* medial lobe not fused to lateral lobe; medial lobe with 2 recurved robust setae, lateral lobe with 1 large recurved robust setae. *Maxilliped* consists of 3 articles; palp article 2 without setae; article 3 with 4 recurved robust setae.

Pereopod 1 basis 1.7 times as long as greatest width; ischium 0.6 times as long as basis; merus proximal margin with slight bulbous protrusion; carpus with rounded proximal margin; propodus 1.3 times as long as wide; dactylus slender, 1.8 as long as propodus, 3.8 times as long as basal width. *Pereopod 7* basis 1.6 times as long as greatest width; ischium 0.9 as long as basis, without protrusions; merus proximal margin with slight bulbous protrusion, 0.6 times as long as wide, 0.3 as long as ischium; carpus 0.6 times as long as wide, 0.2 as long as ischium, without bulbous protrusion; propodus 1.4 times as long as wide, 0.5 as long as ischium; dactylus slender, 1.7 as long as propodus, 3.6 times as long as basal width.

Pleopod exopod larger than endopod. *Pleopod 1* exopod 1.3 times as long as wide, lateral margin weakly convex, distally narrowly rounded, medial margin weakly oblique; endopod 1.6 times as long as wide, lateral margin slightly straight, distally narrowly rounded, medial margin straight; peduncle 0.3 times as wide as long, without retinaculae. *Pleopod 2* appendix masculina with parallel margins, 0.8 times as long as endopod, distally acute. *Pleopod 5* with fleshy folds present. Peduncle lobes present, increasing in size from pleopod 1 to 5.

Uropod same length as pleotelson, peduncle 0.8 times longer than rami, peduncle lateral margin without setae; rami extending to pleotelson apex, marginal setae absent, apices narrowly rounded. 2.5 times as long as greatest width, without setae. *Exopod 2.4* times as long as greatest width, without setae.

Penes prominent, 2.3 times as long as basal width, tubercles connecting at base.

Distribution

Off the Zambezi estuary, Mozambique and Tanjona Vilanandro, north-western coast of Madagascar (previously Cape Saint André) (Rokicki 1982); Mayotte Island (Trilles 1976); Pakistan (Behera et al. 2016); India (Rameshkumar et al. 2013b, 2015) as well as Indian eastern coast and Visakhapatnam (north-west Bay of Bengal) (Behera et al. 2016); Thailand (Nagasawa and Petchsupa 2009) and Ko Khram (Schioedte and Meinert 1884); Indonesia (Milne Edwards 1840; Trilles 1979); China (Yu and Li 2003); Mariveles and Luzon Islands (Schioedte and Meinert 1884; Trilles 1976; Yamauchi et al. 2005) and Panay Gulf, Province of Iloilo, the Philippines (Cruz-Lacierda and Nagasawa 2017); Arafura Sea, off the Northern Territory coast of Australia (Bruce 1990); and north-western Australia (Avdeev 1978).

The only previous record of *N. indica* from Mozambique was by Rokicki (1982), off the Zambezi river estuary. This

paper presents the first report of *N. indica* from Maputo Bay, Mozambique, representing the most southern distribution in the Indian Ocean for this species. From the distribution data, it is evident that most *N. indica* recordings have been made from the eastern regions of the Indian Ocean. The localities from this study, as well as from Trilles (1976) and Rokicki (1982), provide the only evidence of the presence of *N. indica* from the western region of the Indian Ocean. Records of *N. indica* correspond to the distribution pattern of their fish hosts.

Hosts

Hosts are usually pelagic and demersal marine teleosts with a preference towards schooling fish, especially those from the family Carangidae. Hosts include smallmouth scad, *Alepes apercna* Grant, 1987 (see Trilles 1976); Indian mackerel, *Rastelliger kanagurta* Cuvier, 1816 (see Avdeev 1978; Rokicki 1982; Ghani 2003; Rameshkumar et al. 2015); blackfin scad, *Alepes melanopectera* Swainson, 1839 (previously as *Atule malam* Bleeker, 1851) (see Avdeev 1978); bigeye scad *Selar crumenophthalmus* Bloch, 1793 (see Rokicki 1982; Bruce 1990; Nagasawa and Petchsupa 2009; Neeraja et al. 2014; Cruz-Lacierda and Nagasawa 2017; present study); *Herklotsichthys* sp. (see Bruce 1990; Ghani 2003; Yu and Li 2003); and *Decapterus* sp. including the Indian scad, *D. russelli* Ruppell, 1830 (see Ghani 2003). Other recent host records include the pugnose ponyfish, *Secutor insidiator* Bloch, 1787 (see Behera et al. 2016), and the redbait scad *Decapterus kurroides* Bleeker, 1855 (see Cruz-Lacierda and Nagasawa 2017). Yamauchi et al. (2005) obtained *N. indica* from the stomach of the common dolphinfish, *Coryphaena hippurus* Linnaeus, 1758 where the natural host would have been eaten by the dolphinfish.

Behera et al. (2016) recorded *N. indica* from Randall's threadfin bream, *Nemipterus randalli* Russell, 1986, which is a doubtful host record. It is the first and only host record from the family Nemipteridae and a photograph provided by Behera et al. (2016: see fig. 5c) does not represent *N. indica*.

Molecular characterisation

Two ovigerous females were sequenced (forward and reverse). Ovigerous female (30.0 mm TL, 17 mm W) produced a 686 bp contig of the COI gene (accession number: MF628258), and ovigerous female (26.0 mm TL, 13 mm W) produced a 687 bp and 679 bp contig of the COI gene (accession numbers: MF628259 and MF628260), respectively. These COI gene sequences are the first sequences to be generated for *N. indica* and the genus *Norileca*, therefore species validity and generic placement based on molecular evidence could not be done.

Remarks

Norileca indica attaches to the ventral part of the host branchial cavity, with the cephalon to the anterior end of the host, and with its ventral side (abdomen/brood pouch) directed outwards, facing the branchial operculum (Bruce 1990; Neeraja et al. 2014; Rameshkumar et al. 2015; Behera et al. 2016). Its asymmetrical body is twisted to the left when it occupies the right branchial cavity and

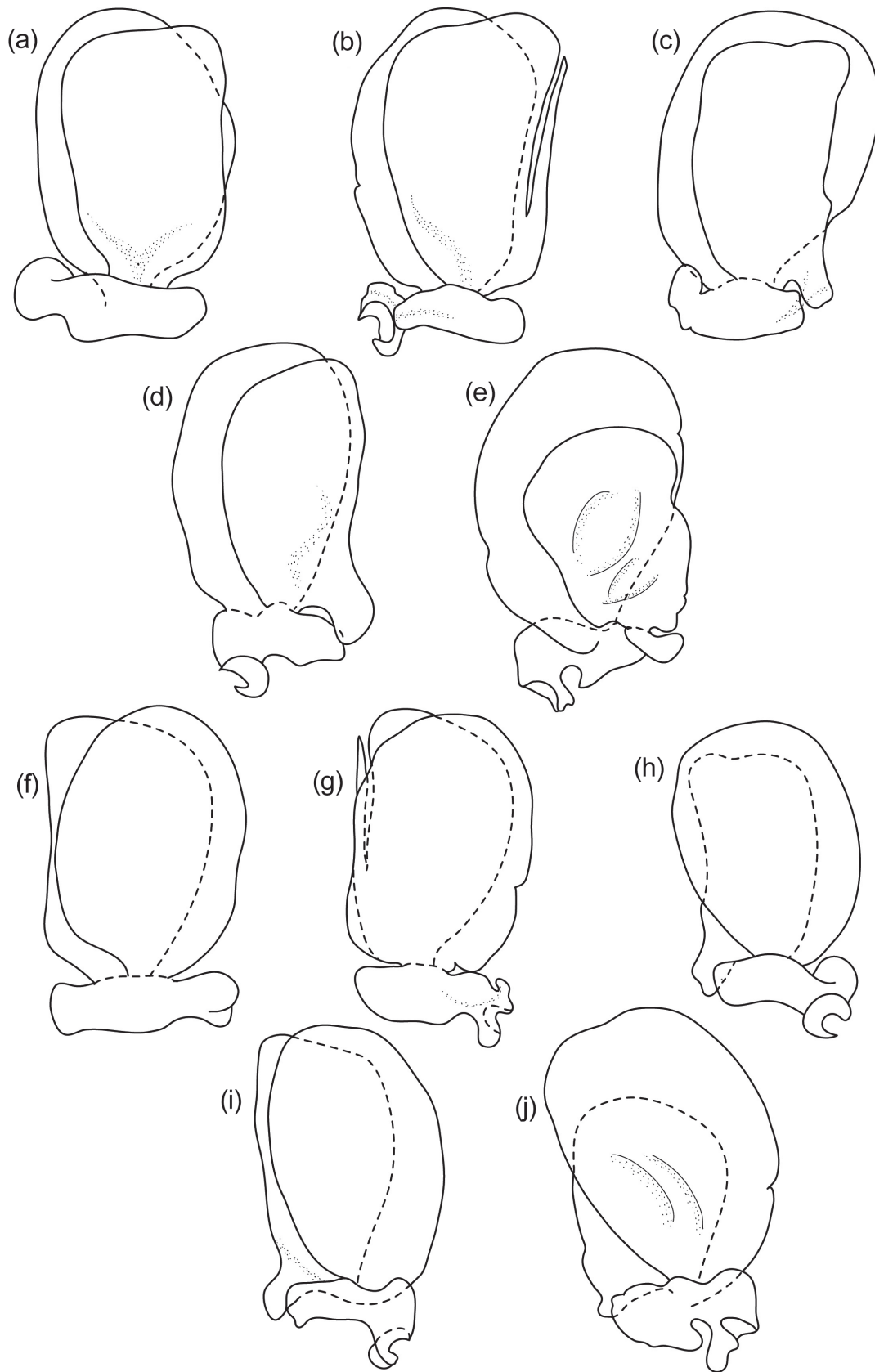


Figure 7: *Norileca indica* (Milne Edwards, 1840) male (11.0 mm TL, 3.0 mm W) (SAMC-A089028). (a) Pleopod 1 dorsal view, (b) pleopod 2 dorsal view, (c) pleopod 3 dorsal view, (d) pleopod 4 dorsal view, (e) pleopod 5 dorsal view, (f) pleopod 1 ventral view, (g) pleopod 2 ventral view, (h) pleopod 3 ventral view, (i) pleopod 4 ventral view, (j) pleopod 5 ventral view

twisted to the right when it occupies the left branchial cavity (Nagasawa and Petchsupa 2009; Neeraja et al. 2014). *Norileca indica* can be recognised by its twisted body, with a 2.1–2.5 length-to-width body ratio and the oval eyes have distinct margins. The pleotelson of *N. indica* is approximately as long as it is wide and the uropods are two-thirds the length of the pleotelson length. Furthermore, pleonite 5 is about as wide as pleonite 1.

Males are similar in appearance to females but smaller in size and with a straight body shape. Ovigerous females differ from non-ovigerous females in having a larger body length-to-width ratio as they become wider as eggs develop in the broodpouch. Both Rokicki (1982) and Bruce (1990) reported that *N. indica* is ventrally positioned in the host gill cavity with the cephalon facing the anterior end of the host and the abdomen outwards toward the operculum (in a lateral position). Four of the currently collected specimens were also ventrally positioned; however, two were positioned with the dorsal surface to the operculum.

Norileca indica can be distinguished from *N. triangulata* by being larger than the latter, with its body twisted to the side, a straight sided pleon, smaller eyes, as well as shorter uropods and a shorter mandible palp article 3 (Bruce 1990). It differs from *N. borealis* by having shorter uropods, larger eyes, and a larger length-to-width ratio of the pleotelson. Pleonite 1 and 5 of *N. indica* are more equal in width than that of *N. borealis* where pleonite 5 is narrower than pleonite 1. In addition, *N. borealis* has a medial lobe with 4 recurved robust setae (two recurved robust setae on *N. indica*), and article 3 of the maxilliped with three recurved robust setae (4 recurved robust setae on *N. indica*) (Javed and Yasmeen 1999). Even though *N. borealis* and *N. triangulata* are more similar to each other than to *N. indica*, they can be distinguished from each other by body shape, ventral margin of the cephalon, as well as pleon and pleopod morphology.

Since the redescription of *N. indica* in 1990, records of this species from the Indian subcontinent have mainly been made due to the collection of its fish hosts for subsistence and commercial use by local fisherman. Many of its host species, including *Selar crumenophthalmus* and *Decapterus kurroides*, are considered to be commercially important fish species (Argente et al. 2014; Cruz-Lacierda and Nagasawa 2017). Other publications from this region provided new host or locality information on *N. indica* as well as some ecological data including prevalence, mean intensity and abundance (Neeraja et al. 2014; Rameshkumar et al. 2015; Behera et al. 2016; Jithin et al. 2016). Despite this species being frequently collected, limited work has been done recently on the morphology and taxonomy of this species.

Discussion

The redescription of *N. indica* by Bruce (1990) was based on non-ovigerous females. Here we present the first detailed redescription of an ovigerous female specimen. Ovigerous females display diagnostic characteristics and structures that may not be present or as well developed in non-ovigerous females and males.

Cymothoid isopods are protandrous hermaphrodites, making them difficult to identify during different developmental stages. As part of the female development, the pleotelson becomes wider and other structures (such as the gonopod, eye- and uropod perimeter, and the first antenna) become shorter in length (Cook and Munguia 2015). Males tend to be morphologically similar to one another; therefore, accurate and comprehensive descriptions of males are essential to ensure species identification can potentially be made in the absence of ovigerous females.

Norileca indica is widely distributed and morphological analyses combined with molecular analyses will provide a better understanding of this species. These analyses will confirm species identity, even during the natatory stage of development (see Jones et al. 2008). It is essential to combine genetic characterisation of a species with an accompanying description of that species based on the same material in order to verify the identification of the species. Only when the identification of the species is accurate can phylogenetic analysis be effective, providing useful information without having the confusion of misidentified species as is currently the problem with cymothoids.

The use of combined molecular and morphological data for phylogenetics is becoming more prominent within taxonomic publications. This combination improves the resolution, internal support and overall quality of phylogenetic studies (Caddick et al. 2002; Scotland et al. 2003). In many instances, there exists a lack of either morphological or molecular data (Giribet et al. 2001). This is also the case with *N. indica*, as no other publication is available that provides a comprehensive data set of both morphological and molecular results of this species. The use of molecular techniques seems to eliminate morphological bias as well as over- and under-estimations of biodiversity that is occasionally associated with traditional morphological analyses (Lefébure et al. 2006).

The first molecular characterisation of *N. indica* presented here contributes to the limited pool of molecular information of the Cymothoidae (currently only 28 of the 385 known and accepted cymothoid species have been sequenced), while also providing a *Norileca* COI sequence that can be used for cymothoid studies as well as species identifications.

Acknowledgements — The project was funded through a Western Indian Ocean Marine Science Association (WIOMSA) Marine Research Grant as well as a Claude Leon Foundation postdoctoral fellowship for KA Hadfield. The financial assistance of the National Research Foundation (NRF) (NRF project IFR170210222411 grant 109352, NJ Smit, PI) and the Foundational Biodiversity Information Programme (DST-NRF FBIP, UID: 108783, S van der Wal) is hereby acknowledged. Opinions expressed, and conclusions arrived at, are those of the authors and are not necessarily those of the NRF. The authors would like to thank Wynand Vlok for assistance in collecting the isopods. This is contribution number 213 from the NWU-Water Research Group.

References

- Aneesh TP, Kappalli S, Kottarathil HA, Gopinathan A, Paul TJ. 2015. *Cymothoa frontalis*, a cymothoid isopod parasitizing the belonid fish *Strongylura strongylura* from the Malabar Coast (Kerala, India): redescription, description, prevalence and life cycle. *Zoological Studies* 54: 42.

- Argente FAT, Narido CI, Palla HP, Celedonio MA. 2014. A review on the biology and parasites of the big-eye scad, *Selar crumenophthalmus* (Bloch, 1793). *Journal of Multidisciplinary Scientific Research* 2: 3–8.
- Avdeev V. 1978. Notes on the distribution of the marine Cymothoidae (Isopoda, Crustacea) in the Australian–New Zealand region. *Folia Parasitologica* 25: 281–283.
- Behera PR, Ghosh S, Pattnaik P, Rao MH. 2016. Maiden occurrence of the isopod, *Norileca indica* (H. Milne Edwards, 1840) in pelagic and demersal finfishes of Visakhapatnam waters along north-west Bay of Bengal. *Indian Journal of Geo-Marine Sciences* 45: 856–862.
- Beumer JP, Ashburner LD, Burbury ME, Jette E, Latham DJ. 1982. A checklist of the parasites of fishes from Australia and its adjacent Antarctic territories. *Technical Communication* no. 48. St Albans: Commonwealth Institute of Parasitology.
- Bleeker P. 1857. Recherches sur les Crustacés de L'Inde Archipelagique. II. Sur les Isopodes Cymothoadiens de L'Archipel Indien. *Natuurkundige vereeniging in Nederlandsche-Indie, Batavia, Verhandelingen* 2: 20–40.
- Borcea I. 1933. *Lironeca pontica* n.sp., parasites des Aloses et sardines de la Mer Noif. *Bulletin du Muséum national d'histoire naturelle, 3e serie* 5: 128–129.
- Brandt A, Poore GCB. 2003. Higher classification of the flabelliferan and related Isopoda based on a reappraisal of relationships. *Invertebrate Systematics* 17: 893–923.
- Bruce NL. 1990. The genera *Catoessa*, *Elthusa*, *Enispa*, *Ichthyoxenus*, *Idusa*, *Livoneca* and *Norileca* n. gen. (Isopoda, Cymothoidae), crustacean parasites of marine fishes, with descriptions of eastern Australian species. *Records of the Australian Museum* 42: 247–300.
- Bruce NL, Lew Ton HM, Poore GCB. 2002. Cymothoidae Leach, 1814. In: Poore GCB (ed.), *Crustacea: Malacostraca: Syncarida and Peracarida: Isopoda, Tanaidacea, Mictacea, Thermosbaenacea, Spelaeogriphacea*. Melbourne: CSIRO Publishing. pp 168–183.
- Caddick LR, Rudall PJ, Wilkin P, Hedderson TA, Chase MW. 2002. Phylogenetics of Dioscoreales based on combined analyses of morphological and molecular data. *Botanical Journal of the Linnean Society* 138: 123–144.
- Coleman CO, Lowry JK, Macfarlane T. 2010. DELTA for beginners: an introduction into the taxonomy software package DELTA. *ZooKeys* 45: 1–75.
- Cook C, Munguia P. 2015. Sex change and morphological transitions in a marine ectoparasite. *Marine Ecology* 36: 337–346.
- Cruz-Lacierda ER, Nagasawa K. 2017. First record of *Norileca indica* (Isopoda, Cymothoidae) parasitic on *Selar crumenophthalmus* and *Decapterus kurroides* (Perciformes, Carangidae) in the Philippines. *Comparative Parasitology* 84: 60–63.
- Eschmeyer WN, Fricke R, van der Laan R (eds). 2017. *Catalog of Fishes: Genera, Species, References*. Available at <https://www.calacademy.org/scientists/projects/catalog-of-fishes> [accessed May 2017].
- Folmer O, Black M, Hoeh W, Lutz R, Vrijenhoek R. 1994. DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology* 3: 294–299.
- Froese R, Pauly D. 2017. FishBase. World Wide Web electronic publication. Available at <http://www.fishbase.org> [accessed May 2017].
- Gerstaecker A. 1882. Sechste Ordnung. Isopoda Asseln (Part). Dr. HG Bronn 'Die Klassen und Ordnung des Thier-Reichs, wissenschaftlich dargestellt in Wort und Bild. *Funfter* 2: 97–278.
- Ghani N. 2003. Isopod parasites of marine fishes of Pakistan. *Proceedings of Pakistan Congress of Zoology* 23: 217–221.
- Giribet G, Edgecombe GD, Wheeler WC. 2001. Arthropod phylogeny based on eight molecular loci and morphology. *Nature* 413: 157.
- Hadfield KA. 2012. The biodiversity and systematics of marine fish parasitic isopods of the family Cymothoidae from southern Africa. PhD thesis, University of Johannesburg, South Africa.
- Hadfield KA, Bruce NL, Smit NJ. 2010. Redescription of the monotypic genus *Cinusa* Schioedte and Meinert, 1884 (Isopoda, Cymothoidae), a buccal-cavity isopod from South Africa. *Zootaxa* 2437: 51–68.
- Hadfield KA, Bruce NL, Smit NJ. 2013. Review of the fish-parasitic genus *Cymothoa* Fabricius, 1793 (Isopoda, Cymothoidae, Crustacea) from the southwestern Indian Ocean, including a new species from South Africa. *Zootaxa* 3640: 152–176.
- Hadfield KA, Bruce NL, Smit NJ. 2016. Redescription of poorly known species of *Ceratothoa* Dana, 1852 (Crustacea, Isopoda, Cymothoidae), based on original type material. *ZooKeys* 592: 39–91.
- Heller C. 1868. Zur naheron Kenntniss der in den sussen Gewassen des sudlichen Europa vorkommenden Meerescrustaceen. *Zeitschrift für Wissenschaftliche Zoologie* 19: 156–162.
- Javed W, Yasmeen R. 1999. A new parasitic isopod of the genus *Norileca* (Crustacea, Cymothoidae) from the Arabian Sea. *Journal of Marine Science* 8: 55–60.
- Jithin K, Swapna A, Kumar RR, Venu S, Helna AK, Sudha K. 2016. Studies on crustacean parasites from commercial marine fish along the Andaman Coast in comparison with Malabar Coast of Kerala of Indian EEZ. *World Journal of Fish and Marine Sciences* 8: 47–53.
- Jones CM, Miller TL, Grutter AS, Cribb TH. 2008. Natatory-stage cymothoid isopods: description, molecular identification and evolution of attachment. *International Journal for Parasitology* 38: 477–491.
- Ketmaier V, Joyce DA, Horton T, Mariani S. 2008. A molecular phylogenetic framework for the evolution of parasitic strategies in cymothoid isopods (Crustacea). *Journal of Zoological Systematics and Evolutionary Research* 46: 19–23.
- Lefébure T, Douady CJ, Gouy M, Gibert J. 2006. Relationship between morphological taxonomy and molecular divergence within Crustacea: proposal of a molecular threshold to help species delimitation. *Molecular Phylogenetics and Evolution* 40: 435–447.
- Milne Edwards H. 1840. *Histoire naturelle des crustacés: comprennent l'anatomie, la physiologie et la classification de ces animaux*, vol. 3. Paris: Roret.
- Nagasawa K, Petchsupa N. 2009. *Norileca indica* (Isopoda, Cymothoidae) parasitic on bigeye scad *Selar crumenophthalmus* in Thailand. *Journal of Biogeography* 11: 131–133.
- Neeraja T, Tripathi G, Shameem U. 2014. Occurrence of the isopod, *Norileca indica* (Isopoda: Cymothoidae) on bigeye scad, *Selar crumenophthalmus* (Bloch) off Mumbai coast, India. *Indian Journal of Fisheries* 61: 49–56.
- Nierstrasz HF. 1915. Die Isopoden-Sammlung im Naturhistorischen Reichsmuseum zu Leiden — 1. Cymothoidae. *Zoologische Mededelingen (Leiden)* 1: 71–108.
- Nierstrasz HF. 1931. Isopoda genuina. II. Flabellifera. In: Weber M, de Beaufort LF (eds), *Die Isopoden der Siboga-Expedition Siboga Expeditie (Uitkomsten op Zoölogisch, Botanisch, Oceanographisch en Geologisch Gebied verzameld in de Oost-Indische 1899–1900 aan boord HM Siboga onder commando van Luitenant ter zee 1e kl GF Tydeman)*. Leiden: EJ Brill. pp 123–233.
- Rameshkumar G, Ramesh M, Ravichandran S, Trilles J-P, Subbiah S. 2015. New record of *Norileca indica* from the west coast of India. *Journal of Parasitic Diseases* 39: 712–715.
- Rameshkumar G, Ravichandran S. 2015. First occurrence of *Norileca triangulata* (Crustacea: Isopoda: Cymothoidae) from Indian marine fishes. *Journal of Parasitic Diseases* 39: 33–36.

- Rameshkumar G, Ravichandran S, Sivasubramanian K. 2013a. Invasion of parasitic isopods in marine fishes. *Journal of Coastal Life Medicine* 1: 99–105.
- Rameshkumar G, Ravichandran S, Sivasubramanian K, Trilles J-P. 2013b. New occurrence of parasitic isopods from Indian fishes. *Journal of Parasitic Diseases* 37: 42–46.
- Richardson H. 1910. Marine isopods collected in the Philippines by the U.S. Fisheries steamer *Albatross* in 1907–1908. *Bureau of Fisheries Document* no. 736. Washington, DC: Department of Commerce and Labor, Bureau of Fisheries.
- Rokicki J. 1982. *Lironeca indica* Edwards, 1840 (Crustacea, Isopoda) from *Selar crumenophthalmus* (Bloch). *Wiadomosci Parazytologiczne* 28: 205–206.
- Schioedte JC, Meinert F. 1884. Symbolæ ad monographium Cymothoarum crustaceorum isopodum familiæ. IV. Cymothoidæ Trib. II. Cymothoinæ. Trib. III: Livonecinæ. *Naturhistorisk Tidsskrift, Kjøbenhavn* 14: 221–454.
- Scotland RW, Olmstead RG, Bennett JR. 2003. Phylogeny reconstruction: the role of morphology. *Systematic Biology* 52: 539–548.
- Trilles J-P. 1976. Les Cymothoidae (Isopoda, Flabellifera) des collections du Muséum national d'Histoire naturelle de Paris. IV. Les Lironecinae Schioedte et Meinert, 1884. *Bulletin du Muséum national d'histoire naturelle, 3e serie, Zoologie* 390: 773–800.
- Trilles J-P. 1979. Les Cymothoidae (Isopoda, Flabellifera; parasites de poissons) du Rijksmuseum van Natuurlijke Historie te Leiden II. Afrique, Amérique et régions Indo-Ouest-Pacifiques. *Zoologische Mededelingen (Leiden)* 54: 245–275.
- Trilles J-P. 1994 [for 1991]. Les Cymothoidae (Crustacea, Isopoda) du Monde. Prodrôme pour une faune. *Studia Marina* 21–22: 1–288.
- Yamauchi T, Ohtsuka S, Nagasawa K. 2005. Ectoparasitic Isopod, *Norileca indica* (Crustacea, Isopoda, Cymothoidae), obtained from the stomach of *Coryphaena hippurus* (Perciformes, Coryphaenidae) in the Philippines. *Journal of Biogeography* 7: 25–27.
- Yu H, Li X. 2003. Study on the Cymothoidae from Chinese waters. *Studia Marina Sinica* 45: 223–238.

